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### Lack of Behavioral Effects in the Rhesus Monkey: High Peak Microwave Pulses at 1.3 GHz

John A. D'Andrea, Brenda L. Cobb, and John O. de Lorge

Bioenvironmental Sciences Department, Naval Aerospace Medical Research Laboratory, Pensacola, Florida

The current safety standards for radiofrequency and microwave exposure do not limit the peak power of microwave pulses for general or occupational exposures. While some biological effects, primarily the auditory effect, depend on pulsed microwaves, hazards associated with very high peak-power microwave pulses in the absence of whole-body heating are unknown. Five rhesus monkeys, Macaca mulatta, were exposed to peak-power densities of 131.8 W/cm<sup>2</sup> (RMS) while performing a time-related behavioral task. The task was composed of a multiple schedule of reinforcement consisting of three distinct behavioral components: inter-response time, time discrimination, and fixed interval. Trained monkeys performed the multiple schedule during exposure to 1.3-GHz pulses at low pulse-repetition rates (2-32 Hz). No significant change was observed in any behavior during irradiation as compared to sham-irradiation sessions. Generalization of these findings to experimental results with higher peak-power densities, other pulse rates, different carrier frequencies, or other behaviors is limited.

Key words: pulsed microwaves, behavior, macaca mulatta

#### INTRODUCTION

At the present time, microwave safety standards [ANSI, 1982; NCRP, 1986] recommend limiting exposure to a whole-body specific absorption rate (SAR) of 0.4 W/kg and a localized tissue peak SAR of 8 W/kg. The standards, however, do not limit the peak power of pulsed microwave fields. Microwave fields with high peak powers but low pulse-repetition rates may satisfy the currently accepted safe SAR limits, but the possibility of adverse health effects has caused some concern in both civilian and military occupational environments. The most widely studied biological effect of pulsed microwaves has been auditory sensation, putatively caused by thermoelastic expansion of brain tissue [Lin, 1978]. Recent studies have investigated the microwave pulse parameters necessary to produce acoustic mechanical vibrations in brain tissue of several mammalian species [Lin, 1980; Olsen and Lin, 1983]. The concern over potential adverse health effects stems not from the relatively low power

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Address reprint requests to Dr. John A. D'Andrea, Bioenvironmental Sciences Dept., Naval Aerospace Medical Research Laboratory, Pensacola, FL 32508-5700.

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microwave pulses necessary to produce auditory stimulation but from very high peak-power pulses capable of producing intense mechanical vibration in brain tissue. This concern appears justified because new devices with very high peak output powers and pulses of short duration are continually being developed.

Recent behavioral studies, designed to determine whether pulsed waves (PW) can facilitate behavioral effects compared to continuous wave (CW) microwaves have, however, produced conflicting results. Thomas et al. [1982] investigated the effect of PW and CW microwaves on a differential-reinforcement-of-low-rate (DRL) schedule at whole-body average SARs of 0.2–3.6 W/kg and peak SARs of 0.2 to 3.6 kW/kg (authors' estimate). The rate of appropriately timed responses by rats on this schedule was consistently disrupted by PW microwaves but not by CW microwaves at SARs of 2.5 and 3.6 W/kg. In contrast, a recent study by Lebovitz [1983] found that rats performing a multicomponent fixed ratio (FR) and timeout (TO) reinforcement schedule were not differentially affected by PW and CW microwave exposure. In this case whole-body average SARs of 5.9 W/kg and 6.7 W/kg were used for CW and PW exposure, respectively. The peak SAR for the PW exposures was 11.2 kW/kg (author's estimate).

Behavioral change, however, has continued to be a sensitive assay for microwave-induced effects. In particular, time-based schedules of reinforcement have proven very sensitive to microwave exposure [Sanza and deLorge, 1977; D'Andrea et al., 1977; deLorge, 1984; Thomas et al., 1982]. A safety standard to govern high-peak microwave pulses can only be established from an extensive base of experimental data. The study reported here is one part of a Navy Department research program to determine thresholds for changes in schedule-induced behavior produced by high peak power microwave pulses.

## MATERIALS AND METHODS Subjects

Five male, juvenile rhesus monkeys (Macaca mulatta) obtained from the NAMRL primate breeding program were used as subjects. The mean body mass of the subjects during the study was 5.2 kg (± 0.23 kg, SEM). The subjects were fed daily a standard primate diet (Wayne Co., 24% protein) in sufficient quantities (freely available in their cages) to produce an animal of normal size and mass for that age and housing condition. Prior to training, the animals were fed a reduced amount of the same diet daily until their body mass was reduced by 5% of the previously determined ad libitum weight. During the course of the experiment, the monkeys were maintained at this weight except for periods where they were again free-fed for several days (5 to 7) to establish a new ad libitum weight. This procedure resulted in healthy, well-conditioned animals that worked adequately on food-reinforced tasks. The animals obtained their daily food ration (Noyes Co., 750-mg monkey formula L pellets) while performing the experiment. Their diet, during the experiment, was supplemented only with fresh fruit. Water was continuously available in the home cage, which was kept in the vivarium under a 12/12-h light-dark cycle (0700 on, 1900 off). The mean ambient temperature of the vivarium was 21.4 °C (± 0.38 SEM) on the days of microwave and sham exposure. The monkeys had served, for approximately 7 months prior to the current study, as subjects in a previous microwave experiment designed to investigate the effects of microwave pulses at 5.6 GHz

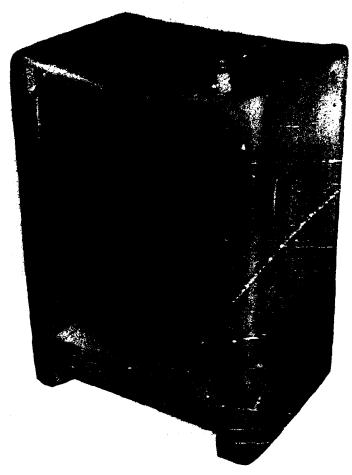


Fig. 1. A rhesus monkey sitting in the Styrofoam restraint chair.

[D'Andrea et al., 1985a]. In that study monkeys were exposed to pulsed microwave energy (0.8  $\mu$ s pulse duration) at pulse-repetition rates of 2, 11, and 16 pps with average whole-body SARs of 0.2–4.41 W/kg. Consistent behavioral effects were not observed in that study.

#### **Apparatus**

Behavioral. Monkeys were restrained in a Styrofoam chair, as shown in Figure 1, and housed within an anechoic chamber which was lined with sheet metal and pyramidal absorbing material (Fig. 2). Two loudspeakers were mounted within the chamber, one to produce white masking noise and the other to produce auditory discriminative stimuli. Four signal lamps (GE No. 751), mounted behind 5-cm<sup>2</sup> translucent Plexiglas panels (blue, white, red, green), were placed 1 m in front of the monkey at eye level and served as visual discriminative stimuli. A pellet feeder, mounted externally on the chamber, delivered food pellets to a depression on

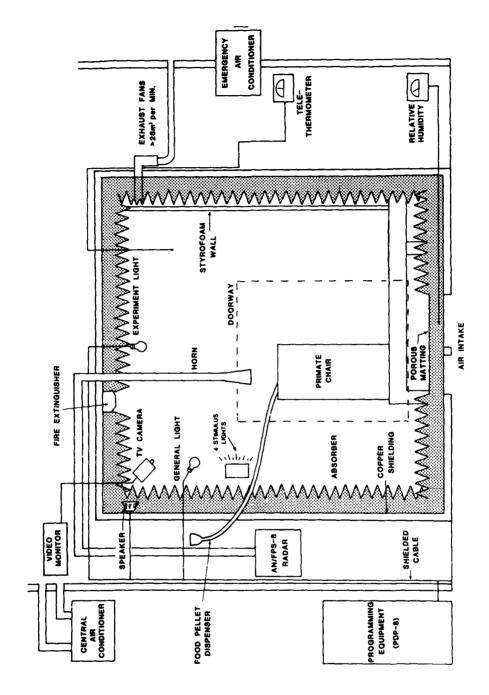


Fig. 2. A schematic representation of the anechoic microwave exposure chamber used to expose monkeys.

top of the chair via a 1.2-m length of Tygon plastic tubing. A 15-W lamp was used for illumination of the chamber, and a closed-circuit TV allowed observation of the monkey during the experiment. A 25-W lamp, mounted inside the chamber, was used to signal the start of an operant session. A background noise level of 72 dBA was measured with an octave analyzer sound-level meter (Bruel & Kjaer, No. 2215) at the location of the Styrofoam chair. The chamber was equipped with a fan that circulated room air through the chamber at 28 m³ per min. A telethermometer (Yellow Springs No. 401) was used to record the ambient temperature within the anechoic chamber; the temperature probe were mounted above the monkey and behind the horn antenna. The mean ambient temperature and relative humidity during microwave and sham exposures were 21.5 °C ( $\pm$  0.14 SEM; range 20.4–23.3 °C) and 34.2% ( $\pm$  5.64%, SEM; range 18–70%).

Exposure system. Pulsed microwave energy was provided by a AN/FPS-8 radar operating at 1.3 GHz. The pulse-repetition rate of the radar was externally controlled by a pulse generator (Hewlett Packard 214A). The radar output was delivered to the chamber via rectangular waveguide (WR-650) which contained a directional coupler (Raytheon No. FA595T) and waveguide switch (RCA No. SA-340/SPS-12). A standard gain horn antenna was mounted vertically 13 cm above the monkey's head. Average power density measurements were made in the near-field at the location normally occupied by the monkey's head using an isotropic probe (Narda 8323) with the radar operating at 150 pps. Average power at the lower pulse rates was extrapolated from this measured value. Peak microwave pulse power was monitored at the directional coupler with a crystal detector (Hewlett Packard No. 423B) and oscilloscope (Tektronix No. 7633). A minicomputer located in an adjacent room controlled events during the experiment and recorded the data.

#### **Procedure**

Behavioral training. After the monkeys reached 95% of free-feeding body weights, they were trained to operate three levers for food pellet reward. The behavioral task consisted of a multiple schedule performed during a 60-min session. Typical cumulative records from such a session are shown in Figure 3, with cumulative responses for the three components (DRL, TD, FI) shown in the upper half and correct responses during the TD component (see below) shown in the lower half. Each of three 10-min components (behavior test schedules) occurred twice during each session. The first component utilized a variation of an inter-response time (IRT) schedule (also known as a differential-reinforcement-of-low-rate schedule, DRL). This schedule required the monkey to withhold responding for at least 8 s but no longer than 12 s. During this 4-s limited hold (LH) "window," two responses on the left lever spaced less than 2 s apart (IRT < 2) produced a food pellet. Inappropriately timed responses did not produce a food pellet and reset the schedule. The nomenclature of the schedule might be identified as a DRL 8 LH4 IRT < 2. A continuous 1,070-Hz tone accompanied the DRL-schedule component. The second component was a time discrimination (TD) schedule where a center-lever response in the presence of a blue lamp produced white light of either short (1, 2, or 3 s) or long (8, 9, or 10 s) duration. Following this randomly chosen duration, the white light was extinguished, and two lights, red and green, were illuminated. If the right lever was depressed following the short white light, a food pellet was delivered. Accordingly, if the left lever was depressed following a long white light, a food pellet was given.

- 10 Minutes -

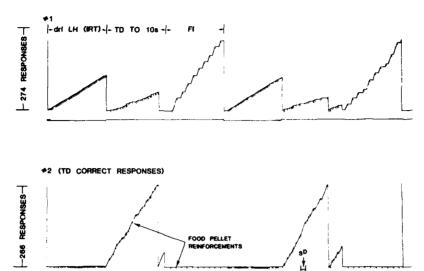


Fig. 3. A typical cumulative record showing responses made during the three schedule components—differential reinforcement of low rates (drl), time discrimination (TD), fixed-interval (FI) (top half)—and correct responses emitted during TD performance (bottom half).

Incorrect responses produced a 10-s time-out period (10 s). The third component, accompanied by a continuous 2,740-Hz tone as a discriminative stimulus, was a fixed-interval schedule (FI) in which the monkey's first response on the right lever after 55 s resulted in the delivery of a food pellet. Approximately 60 baseline sessions were given before the start of sham or microwave sessions (5 days per week  $\times$  12 weeks).

Microwave exposure. Behavioral performance was considered stable when the mean number of responses emitted during each component of a baseline session varied 10% or less than the same components on a previous days session. Following the development of stable behavioral performance, the monkeys were exposed in the k||L orientation, with the electric field traversing ear-to-ear, to 3-µs pulses at pulse-repetition rates and field-power densities shown in Table 1. A repeated-measures experimental design was used where the order of the various exposures and a sham exposure were given randomly for each monkey. Microwave exposures were given one session per week for 60 min while the monkey performed the behavioral task. Each monkey was given one exposure to each of the pulse repetition rates listed in Table 1. The exposures were generally given on Wednesday of each week. During baseline sessions the radar was placed in the standby mode of operation. During a sham-exposure session, the radar was operational but the energy delivered to the waveguide was redirected from the antenna using the waveguide switch to a waveguide resistive load located outside the anechoic chamber.

**Dosimetry.** An estimate of the local SAR in the center of the monkey head was determined using two bag monkey models (5.05 and 5.30 kg), similar to that used by Olsen [1984], and filled with simulated muscle tissue [Chou et al., 1984]. A monkey

TABLE 1. Field-Power Density and Specific Absorption Rate

	Pulse repetition rate (Hz)							
	2	4	8	16	32			
Power density								
Average (mW/cm <sup>2</sup> )	0.92	1.85	3.70	7.40	14.80			
Peak (W/cm <sup>2</sup> )	131.80	131.80	131.80	131.80	131.80			
SAR (W/kg)								
Whole body	0.05	0.10	0.20	0.40	0.80			
Head	0.09	0.18	0.36	0.72	1.44			
Peak SAR (W/kg)								
Whole body	8.3	8.3	8.3	8.3	8.3			
Head	15.0	15.0	15.0	15.0	15.0			

model was mounted in the Styrofoam chair and placed in the anechoic chamber. A small plastic cannula was inserted to the center of the monkey model head (head diameter = 7 cm). A microwave-compatible temperature probe (Vitek No. 101) was then inserted into the cannula. Temperature of the model was then recorded at 30-s intervals before, during, and after microwave exposure. If the temperature remained stable for several minutes then a 90-s microwave exposure was given with the radar operating at 150 pps, producing an average power density at the location of the model of 69.4 mW/cm<sup>2</sup> as measured by a field probe (Narda No. 8323). Each model was exposed three times allowing head temperature to stabilize between each exposure. The local SAR was then calculated using the following formula: SAR (W/kg) =  $c\Delta T/t$  where  $\Delta T$  is the temperature change in degrees Celsius, c is the specific heat in J/kg/°C, and t is the exposure time in seconds.

The whole-body SAR was estimated following procedures given by D'Andrea et al. [1985]. Fresh monkey models were individually irradiated and then quickly placed in a gradient layer calorimeter (Thermonetics No. 2401). The whole-body SAR was determined from heat released by the model while in the calorimeter and compared to a calibrated standard output for the calorimeter. This procedure was repeated four times with freshly filled models. Local SAR and whole-body SAR values at the lower pulse repetition rates were extrapolated from these measured values.

A measure of the mechanical vibration induced in brain tissue by each microwave pulse was accomplished by inserting a hydrophone transducer (International Transducer Corp. No. ITC 1056) into the center of the cranial cavity of the rhesus monkey carcass, that was seated in the Styrofoam chair. The transducer was inserted 4 cm into brain tissue through a small hole placed in the skull overlying the occipital cortex, with the long axis of the hydrophone aligned parallel to the magnetic field. Response of the hydrophone during microwave exposure was monitored with an oscilloscope (Tektronix No. 7633) and compared to a calibration standard supplied with the hydrophone.

#### RESULTS

The estimated power densities and the SARs of the head and the whole body are shown in Table 1. A mean whole-body SAR value of 0.05 W/kg (± 0.01 W/kg,

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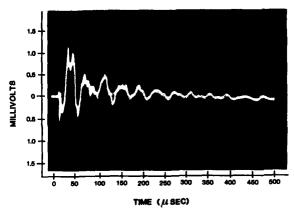


Fig. 4. A typical response of the hydrophone inserted into the center of the cranial cavity of a rhesus monkey carcass and exposed to pulsed microwave radiation.

SEM when normalized to 1 mW/cm<sup>2</sup>) was nearly identical with previous analytical estimates [Durney et al., 1978]. An estimate of the total energy delivered per pulse to the head was 280 mJ/kg; a measure of the induced mechanical pressure in brain tissue using the hydrophone was 84.4 pascals per pulse. The predominant frequency of the waveform was 25 kHz, which is consistent with the shape of the rhesus skull and the sonic conduction in brain tissue [Olsen and Lin, 1983]. A typical hydrophone response during microwave exposure is shown in Figure 4.

The test session conducted the day just prior to each microwave or sham treatment was used as baseline. The mean number of correct responses per minute for each schedule component during microwave and sham exposure sessions were expressed as percentage of the correct responses during baseline sessions. Mean correct responses per minute for the DRL components are shown in Figure 5. A slight but statistically insignificant reduction from baseline performance was observed for all treatments, including the sham condition. The mean correct responses per minute for the TD components are shown in Figure 6. No change from baseline for this dependent measure was observed. The mean correct responses and the mean post reinforcement pause times for the FI components are shown in Figures 7 and 8. respectively. An insignificant departure from baseline performance was observed for the 2-Hz exposures on both measures of FI performance. A one-way analysis of variance for repeated measures (ANOVA) was used to test for microwave effects on each of the dependent measures [Kepple, 1973]. The results of the ANOVAs showed no significant effects (P > .05) due to the microwave treatments on any of the measures.

#### **DISCUSSION AND CONCLUSIONS**

The pulsed-microwave radiation exposures used in this study (131.8 W/cm<sup>2</sup> RMS peak power) did not produce statistically significant alterations of a complex trained behavior in rhesus monkeys. The microwave absorption produced by the irradiation (280 mJ/kg per pulse) was well above the threshold for the auditory

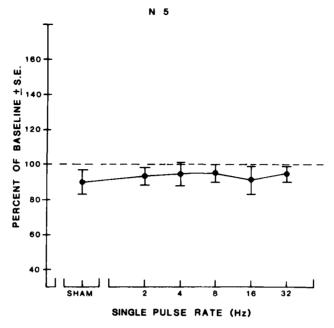


Fig. 5. Mean correct responses per minute as a percentage of baseline performance for the DRL-schedule components for sham and 2- through 32-Hz microwave exposures.

hearing phenomenon [Lin, 1978]. If auditory stimulation occurred in the monkeys, it produced no obvious effect on the trained behavior.

The whole-body SARs produced by the exposure parameters used in this study (0.05-0.8 W/kg) were well below the threshold (4.5 W/kg) found necessary by de Lorge [1984] for disruption of operant performance in rhesus monkeys exposed to 1.3-GHz pulsed microwaves. The peak power of pulses used by de Lorge [1984] was less, 51.4 W/cm² (109 mJ/kg per pulse), compared to 131.80 W/cm² (280 mJ/kg per pulse) used in this study. Nevertheless, the almost threefold increase in peak power and peak absorbed energy used in this study did not compensate for the low average whole-body SAR and did not alter behavior. While significant effects were not observed in this study, the generalization of these findings to higher peak-power densities, other pulse rates, different carrier frequencies, as well as other behaviors, is unwarranted.

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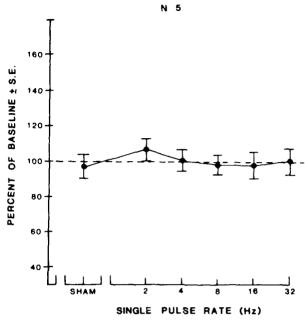


Fig. 6. Mean correct responses per minute as a percentage of baseline performance for the TD-schedule components for sham and 2- through 32-Hz microwave exposures.

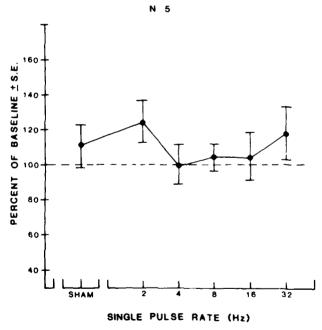


Fig. 7. Mean correct responses per minute as a percentage of baseline performance for the Fl-schedule components for sham and 2- through 32-Hz microwave exposures.

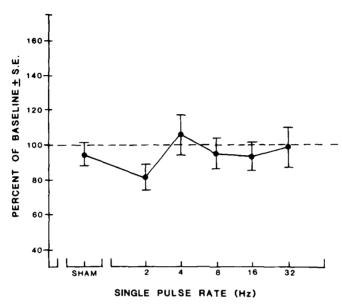


Fig. 8.—Mean post-reinforcement pause times as a percentage of baseline performance for the FI-schedule components for sham and 2- through 32-Hz microwave exposures.

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The animals used in this study were handled in accordance with the *Guide for the Care and Use of Laboratory Animals* prepared by the Committee on Care and Use of Laboratory Animals of the Institute of Laboratory Animal Resources, National Research Council, DHEW, NIH Publication No. 85-23, 1985, and the Animal Welfare Act of 1966, as amended.

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